COMPARATIVE ANALYSIS OF LIPIDS FROM HEN AND QUAIL YOLKS

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Abstract: Structural lipid compounds of hen and quail egg yolks were extracted by solvent extraction procedure according to Folch method. The amount of triglycerides, lecithin and cholesterol which were extracted was to be 18.35, 10.73, 4.66 g/100g yolk for wet hen yolk and 19.93, 7.51, 4.38 g/100g yolk for wet quail yolk. Triglycerides extracted from the yolk look like a solid mass, with substantially white color, non-malleable consistency, that are estimates as 18.35 g/100 g wet hen yolk and 19.93 g/100 g wet quail yolk. Hen egg yolk lecithin has emulsifying qualities more evident than in quail egg yolk lecithin. Lecithin extracted from hen egg yolk formed the milk creaminess structures with high stability, where agglomerations are well fixed and distributed uniformly.

Keywords: egg yolk, lipidomics, triglycerides, lecithin, cholesterol, spectral characteristics, technological properties, instrumental testing.

Introduction

Egg lipids are concentrated in yolk. 70% of the dry substances of egg yolk are represented by lipids. In general the yolk of various strains of hen eggs is 30% to 33% lipid comprising about 65% triglycerides, 28.3…30% phospholipids, 4.0…5.2% cholesterol and 1% other lipids [4, 7, 10].

Triglycerides of egg yolk have the following fatty acid in composition: myristic (14:0), palmitic (16:0), stearic (18:0), oleic (18:1), linoleic, 52.4% (18:2) and clupadonic acid (22:5).

The total amount of saturated fatty acid is approximately 40% of the fatty acids. Major unsaturated fatty acids are oleic, linoleic, and linolenic with a small quantity of $C_{20}\omega6$, $C_{22}\omega6$ and $C_{22}\omega3$ polyunsaturated fatty acids [4].

The polar lipids (namely phospholipids, PL) are represented by several classes: lecithins (79%), cephalins (17%), sphingomyelins (2.5%) and others. The main components of egg-yolk lecithins are phosphatidylcoline (PC) with a presence of 80.5%, and phosphatidylethanol-amines (PE) – 11.7%. Phosphatidylcoline (PC) is the major component of phospholipids of egg yolk (66.7% of total phospholipids), that is more than phospholipids in soybeans (only 33.0%) [7].

The extraction of total lipids and lecithin from yolks is desirable because of the unique properties and valuable applications of these products.

In recent decades lecithins have been developed for use in a wide variety of products including chocolate, confectionery and bakery preparations, dairy products and low fat products [7, 11].

Both lecithin and cholesterol have multiple physiological functions. Lecithin is responsible for elasticity and integrity of vascular and cellular membranes. Cholesterol contributes to the synthesis of steroid and sexual hormones [6].

In some studies was established that solvent composition had a small effect on the extraction of predominant lipid classes (triacylglycerides, cholesterol esters, and phosphatidylcholines). In contrast, extraction of less abundant lipids

(phosphatidylinositols, lyso-lipids, ceramides, and cholesterol sulfates) was greatly influenced by the solvent system used [8].

For the removing the main classes of lipids from animal tissues are using the solvent system chloroform:methanol (2:1, v/v) or Folch method [3]. For the removing the secondary lipidic compounds is required a two-phase system, where initially is preparing a mixture of chloroform:methanol:water, which after mixing, is separated into two layers with the following composition: the upper layer - 3:48:47 and the lower layer - 86:14:1, with high extractability for apolar lipids.

Overall, the Folch method is most effective for the extraction of a broad range of lipid classes, although the hexane-isopropanol method is best for apolar lipids and the MeOH-TBME method is more suitable for lactosyl ceramides [8].

Materials and methods

The research was conducted for egg yolk obtained from hen eggs produced by hens species *Leghorn* on poultry ferm Valea Perjei, Taraclia and *Japanese* quail from SRL Cristilmar, Ungheni. For each sample used in the investigation the average mix was obtained from six egg yolks from a homogeneous batch of eggs. The yolk acidity was determined by titration with NaOH 0.1N, being used phenolphthalein as indicator.

The total lipids were extracted by the Folch method, the solvent used was a mixture of chloroform:ethanol (2:1). Yolk proteins were previously hydrated with sol. NaCl, 0.9%, denaturated by treatment at 65-70 °C for 15-20 min. and then proteins have been removed by centrifugation at 3000 rpm for 10 min. [4]. For the separation of major lipid classes - triglycerides, phosphatidilcoline and cholesterol have been used different solvent systems. So, were separated triglycerides (with ethyl alcohol) and lecithin from lecithin and cholesterol mixture with ethyl alcohol:cyclohexane (2:1) solvent. For better separation of cholesterol, which is contained in the yolk plasma, there was used a pectin gel, 1%.

For the quantitative assay of cholesterol have been used certain identification and quantification reactions, Lieberman-Burchard test [5]. Spectral characteristics of preparations of the major classes of lipids of yolk were recorded in the range 190-1000 nm, using for this the UV-VIS spectrometer, model Hach Lange DR 5000.

Statistical analysis on the extractability and proportions between the classes of lipids was performed using ANOVA simultaneous component analysis (ASCA) [9].

Results and discussions

Phisico-chemical characteristics of egg yolk. Hen eggs weight 66.30±3.27 g (weight category L [1]), while egg yolk was 17.18±1.35 g. Quail eggs weight 11.90±0.62 g, while egg yolk was 4.34±0.78 g. Vitteline index was 0.43 for hen egg yolk and 0.35 for quail egg yolk, which shows the presence of higher surface-performing forces and interactions in hen egg yolk than in quail. Acidity of hen egg yolk was of 12.5 degree of acidity, while of quail egg yolk – 10.3, quail egg yolk is less acid than hen yolk. The intensity of the yellow color was estimated to 5 units for hen yolk and 8 units for quail yolk (according to Roche Fan Yolk scale). Dry substances are forming 48.03±2.87 % by weight for hen yolk, and 46.06±1.59 for quail egg yolk. Yolk:albumen ratio was higher for quail yolk (1/1.28 opposite 1/1.94).

Extraction of total lipids from yolk. Yolk represents an oil-in-water emulsion with very thin structure [2]. Many lipids of yolk formed complex structures with proteins

or carbohydrates. Therefore it is relatively deficient to separate lipids with a high yield and in a pure form. The prior preparation of the sample was initial by the hydration of yolk protein in saline solution, then denaturation of them at moderate temperatures of native, factors that increased the breaking of links between proteins and lipids. The amount of total lipid extracted from egg yolks was higher for hen yolk than in quail yolk (Figure 1).

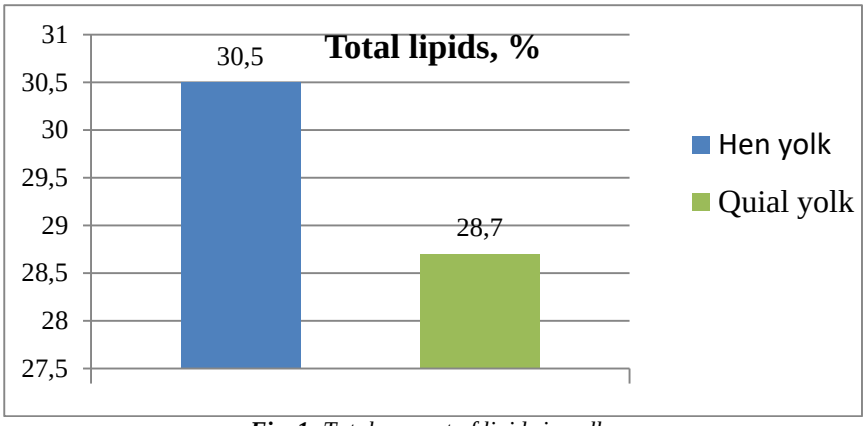

Fig. 1. *Total amount of lipids in yolks*

Separation of major lipids classes. Using a few mixtures of solvents was allowed the effect of separation of major fractions of triglycerides, and then was carried out the separation of cholesterol from lecithin. Triglycerides extracted from the yolk look like a solid mass, with substantially white color, non-malleable consistency, possessing an acidity number equal to 2.0 mg KOH /100 g fats (Figure 2). The lecithin fraction was present in the form of a solid mass of malleable consistency, with strong yellow color.

Fig. 2. *Neutral fats, triglycerides extracted from yolk fats*

Cholesterol, which has a low molecular weight, was separated at last, experimental sample being treated at low temperatures. Quantitative data on the presence of egg yolk lipid fractions are shown in Table 1.

Table 1. Lipid fractions of egg yolk

Lipid fraction	Hen yolk		Quail yolk	
	% of total lipids	g/ 100g yolk	% of total lipids	g/ 100g yolk
Triglycerides	60.16±1.28	18.35±0.45	69.44±1.14	19.93±0.36
Phospholipids	35.18±0.47	10.73±0.17	26.18±0.13	7.51±0.08
Cholesterol	4.66±0.06	1.42±0.03	4.38±0.04	1.26±0.02

Spectral characteristics of the major lipids classes. The UV-VIS absorption spectrum of solution of lecithin in chloroform shows a few peaks shaped as a cascade in the wave band of 250-350 nm, and a shoulder peaks between 400-550 nm (Figure 3).

Experimental investigation for the test samples with cholesterol shows that the absorption spectrum in UV-VIS has the following absorption peaks: at 240 nm (A=2.451), 250 nm (A=2.583), 416 nm (A=0.306). There is a shoulder peak between 450 and 500 nm, whose central wavelength is 460 nm. In actual testing practice absorption peak at 416 nm is used to determine the cholesterol concentration [2].

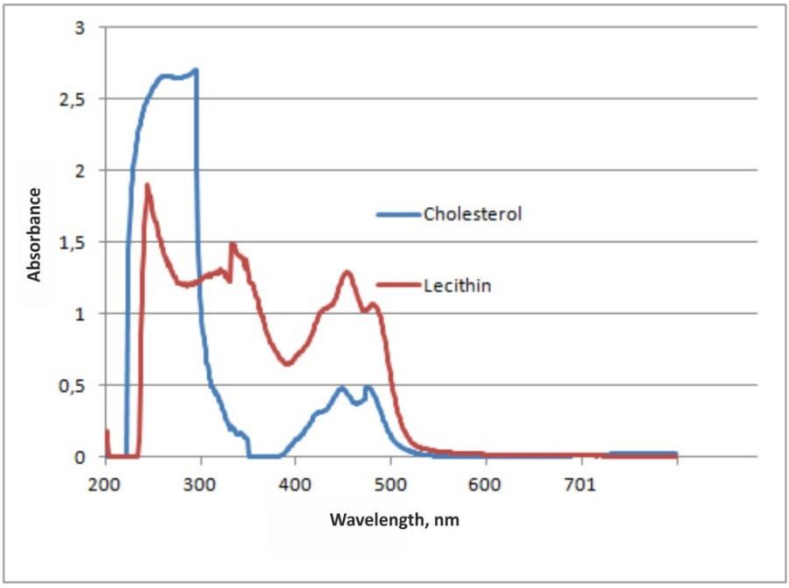

Fig. 3*. Absorbance profile of cholesterol and lecithin in the range of 200-700 nm.*

Technological properties of lecithin from yolk. Lecithin extracted from hen egg yolk was used as a foaming agent in the technology of preparing of the whipped cream. Egg yolk lecithin and milk β-casein are two classes of compounds that have a strong tendency at the air-liquid interface in food creaminess structures. Lecithin extracted from

hen yolk denoted a stronger property for the formation, stabilization and better rheological qualities of milk creaminess structures, which is due to the ability of lecithin to displace β-casein from the cream droplet surface.

Conclusions

1. The egg yolk is a source of multiple compositional ingredients with high technological and nutritional value.

2. Triglycerides extracted from the yolk look like a solid mass, with substantially white color, non-malleable consistency, that are estimates as 18.35 g/100g hen yolk and 19.93 g/100g quail yolk.

3. Both lecithin and cholesterol have a representative profile of UV-Vis spectrum.

4. Hen egg yolk lecithin has emulsifying qualities more evident than in quail egg yolk lecithin.

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